

HAPLOID AND DIPLOID POLLEN IN HYPERICUM PATULUM

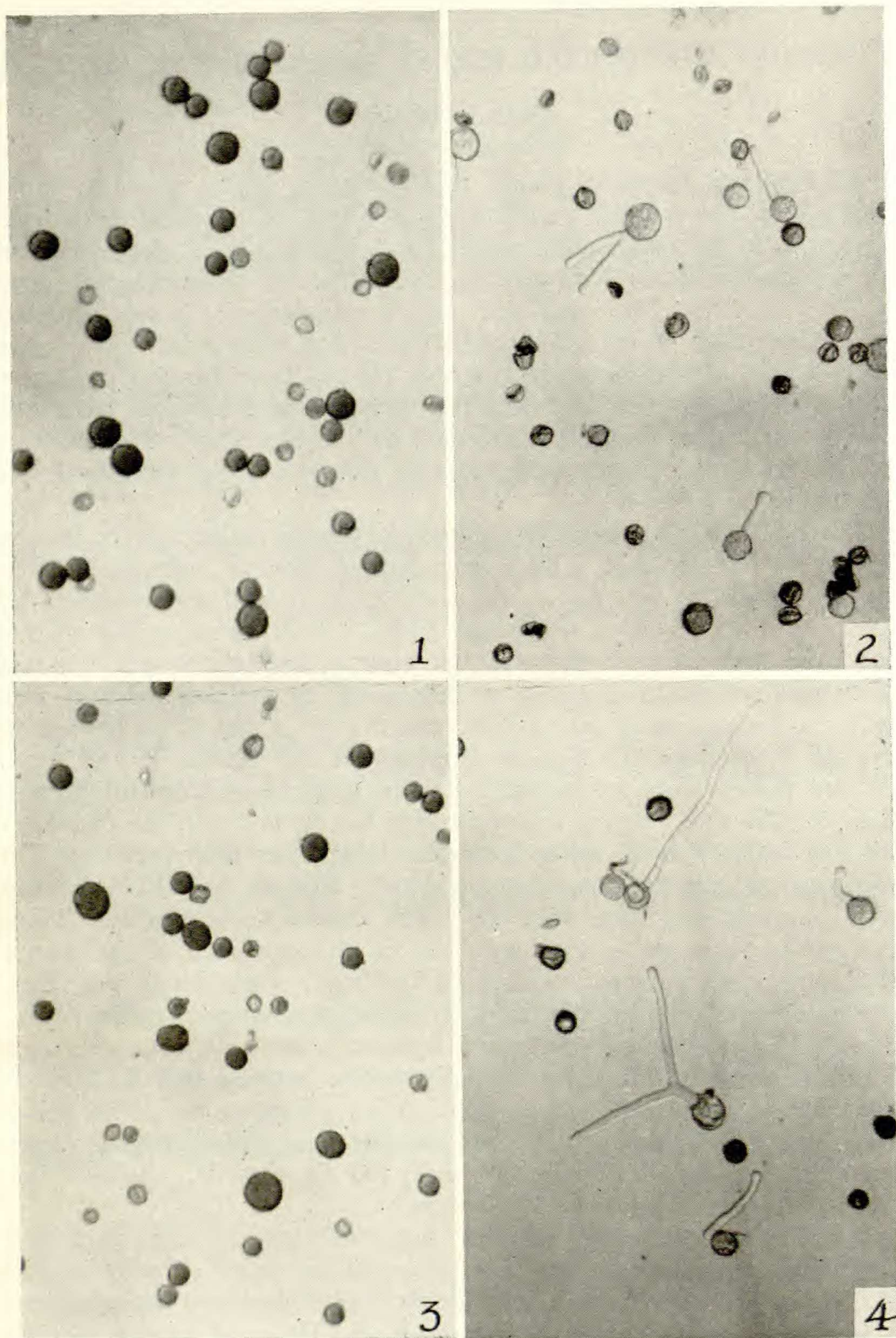
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HYPERICUM PATULUM Thunb. is a spreading shrub, native to Japan and China, but frequently cultivated elsewhere. It is evergreen or semi-evergreen in the southeastern states, but is killed to the ground each year in the Boston area. Even in areas in which it is winter-killed it grows to be 3 to 4 feet high each year, with a rounded growth habit. Cymes of golden yellow flowers $1\frac{1}{2}$ to 2 inches across appear throughout July and August. *Hypericum patulum* var. *Henryi* Bean has larger flowers, 2 to $2\frac{1}{2}$ inches across, and from this variety several cultivars have been derived, including 'Sungold', 'Hidcote', and 'Golden Cup'. The cultivars have even larger, usually darker colored flowers and are more profusely blooming.

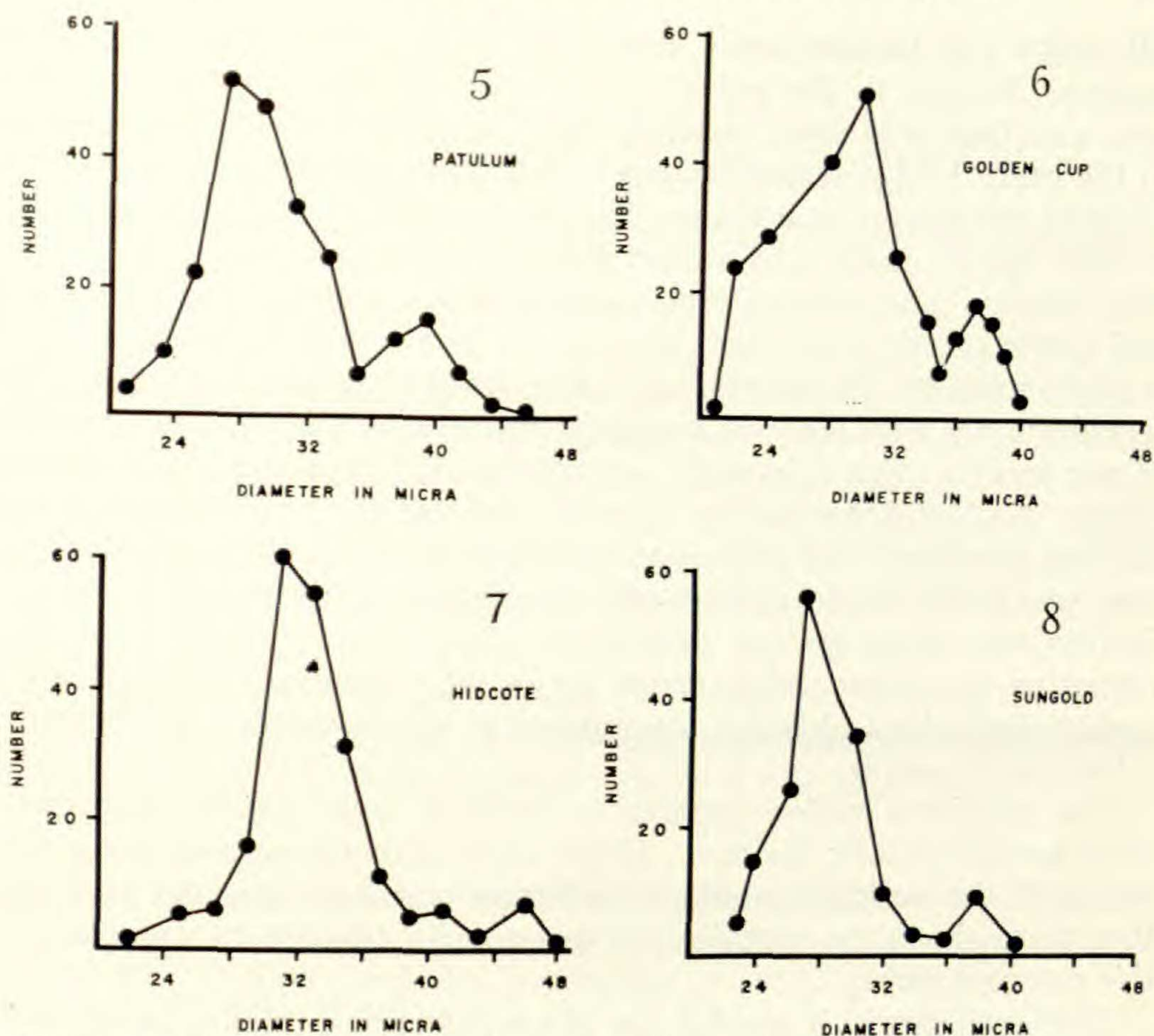
Polyploidy was suspected and a study of mature pollen from herbarium specimens revealed both a high percentage of abortive grains and an unusually large size variation in those appearing normal and staining normally with acetocarmine (FIGURES 1 and 3). Subsequent studies of pollen from buds of fresh material just prior to anthesis showed that the percentage of abortive grains, as determined by staining reaction with either acetocarmine or cotton blue, varied from a low of 30 percent in typical *H. patulum* to a high of 69 percent in the cultivar 'Golden Cup'. Size frequency plots for normally staining grains are shown in FIGURES 5 to 8. The size range is unusually high in this material, the diameters varying from 18 to 45 micra for grains taken from a single anther. In each case the size frequency curve is slightly bimodal, and although there were intermediates, there were two fairly obvious size categories. These are readily apparent in FIGURES 5 to 8. Although each of the graphs shown was made using measurements taken from a single flower, more than one anther was used in order to obtain a larger number of pollen grains. The pattern is essentially the same, however, when pollen from a single anther is measured. Samples from *Hypericum patulum* and *H. patulum* 'Hidcote' were examined using a single anther. Dividing the pollen grains into three groups, large, small, and abortive, the following counts were made; *H. patulum*: 165 large, 378 small, 547 abortive; *H. patulum* 'Hidcote': 111 large, 203 small, 679 abortive.

Germination tests, using a weak sugar solution, revealed that both size groups germinate, apparently with equal frequency (FIGURES 2 and 4). Moreover, pollen tube growth for the first three hours following germination seemed to proceed at approximately the same rate in the different size categories, although measurements of tube length were made on only a small number of cells.

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FIGURES 1-4. Pollen grains from living material of *Hypericum patulum* collected approximately 24 hours before anthesis. FIG. 1, *Hypericum patulum* 'Hidcote'. FIG. 2, *Hypericum patulum*. FIG. 3, *Hypericum patulum*. FIG. 4, *Hypericum patulum* 'Sungold'. Photographed at an initial magnification of approximately 430 \times . Further explanation in text.



FIGURES 5-8. Size frequency plots for pollen grains which showed a normal staining reaction with acetocarmine. FIG. 5, *Hypericum patulum*. FIG. 6, *Hypericum patulum* 'Golden Cup'. FIG. 7, *Hypericum patulum* 'Hidcote'. FIG. 8, *Hypericum patulum* 'Sungold'.

A chromosome number of $2n = 36$ was reported by Sugiura (1936) for *Hypericum patulum*, from material cultivated in a botanical garden. No other counts for this species were reported by Robson and Adams (1968) in a recent survey of chromosome numbers in *Hypericum* and related genera. In the present study chromosome counts and pollen measurements were made on each of the varieties and cultivars listed in TABLE I. All counts were made from plants in the living collection of either the Arnold Arboretum, Harvard University (AA), or the University of Alabama Arboretum (UA). Column 4 in the table indicates the source from which the material was obtained by the respective Arboretum. Unfortunately, no material from native populations was available for study.

TABLE I

<i>Hypericum patulum</i>	$n = 18$	AA 149-49	Kohankie Nursery
<i>H. patulum</i> var. <i>Henryi</i>	$n = 18$	AA 160-45	Kew Gardens, England
<i>H. patulum</i> var. ?	$n = 18$	AA 747-49	Glasnevin, Ireland
<i>H. patulum</i> 'Golden Cup'	$n = 18$	UA 73-67	Neosho Nursery
<i>H. patulum</i> 'Hidcote'	$n = 18$	AA 263-51	Wayside Nursery
<i>H. patulum</i> 'Sungold'	$n = 18$	UA 41-67	Interstate Nursery

All counts and measurements were made from acetocarmine squashes of anthers. Meiosis in the pollen mother cells showed irregularities, however, and there was some variation in chromosome number from one cell to the next. Bridge formation was of frequent occurrence in most of the material examined. In addition, the small and very sticky chromosomes in this species make this rather difficult cytological material. Pairing was irregular and multivalent formation was common. Both trivalents and quadrivalents were seen, with up to four multivalents occurring in a single nucleus. Univalents, too, were observed rather frequently. The univalents lag noticeably at anaphase and usually lead to the formation of micronuclei. This apparently accounts for the large number of abortive grains. Certain other pollen mother cells fail to undergo the second division of meiosis and form dyads rather than tetrads. These cells, therefore, retain the diploid chromosome complement which probably accounts for the very large grains. Preliminary observations indicate that dyad formation and giant pollen grains occur with about equal frequency in a given individual, although the number of dyads counted represented a rather small sample.

The remaining pollen apparently develops from tetrads following a more normal meiotic division. These occur with the greatest frequency, and with the various meiotic disturbances observed it is not surprising that there should be considerable variation in the size of the grains in this category also.

Additional work is needed on these interesting plants, particularly studies of megasporogenesis, and controlled pollination experiments. It should be particularly interesting to make cytological studies of this material from native populations. Although $2n$ pollen seldom competes successfully with $1n$ pollen (Burnham 1962) it is apparent that functional $2n$ pollen is produced by these plants in significant quantities.

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